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Two complexes at the site of microtubule nucleation

LI ShunDai

Department of Biochemistry and Molecular Biology, Pennsylvania State University, University Park, PA 16802, USA

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Cortical microtubule (MT) arrays are dynamic filamentous structures that are essential for cell differentiation and development in plants. However, the molecular mechanisms that control the organization of cortical MT arrays are not well understood. Early studies have revealed that the formation of cortical MT arrays involves MT nucleation on existing cortical MTs. The growth of new MTs follows the polarity of existing MTs and the orientation of new MTs is either in parallel with extant MTs or at a small angle (about 40 degree) to the extant MTs [1]. Nucleation machinery appears to be conserved between animals and plants in which γ-tubulin ring complexes (γ-TuRC) are associated with existing MTs and define the location where MT nucleation initiates [2]. The conservation between animals and plants also extends to another complex, known as the augmin complex, which plays a critical role in the assembly of MT arrays of the spindle and phragmoplast during mitosis [3]. A recent study has shown that both augmin and γ-TuRC are required for the formation of branched MTs in *Xenopus* [4]. Could the augmin complex and γ-TuRC also coordinate MT-dependent MT nucleation in interphase plant cells to orchestrate the formation of cortical MT arrays? In this study, Liu et al. [5] addressed the above question using both genetics and cell biology in interphase plant cells.

By tracking the dynamics of two green fluorescent protein (GFP)-tagged augmin subunits AUG3 and AUG7, the authors showed that the augmin complex was frequently recruited to interphase cortical MTs and often preceded both branching and parallel MT nucleation. The dynamics of AUG3/7-GFP resembles that of γ-TuRC labeled by GCP2-3xGFP. These observations prompted the authors to resolve the co-localization of these two complexes at both temporal and spatial resolution. To investigate the relationship between augmin and γ-TuRC, the authors generated transgenic lines bearing AUG1-2xTagRFP and GCP3-GFP under the control of their native promoters. Co-localization between AUG1-2xTagRFP and GCP3-GFP was extensive, above 80 percent. However, the residence time of augmin was shorter than that of γ-TuRC and augmin appeared at MT nucleation sites earlier than γ-TuRC, causing the authors to propose that the augmin complex recruits γ-TuRC to initiate MT-dependent MT nucleation. Consistent with the proposed function of augmin in initiation of MT-dependent MT nucleation, the frequency of both parallel and branching nucleation events was reduced in augmin knockdown lines in which an artificial microRNA (amiRNA)-AUG6 was expressed.

This study provides convincing evidence that augmin regulates the MT-dependent MT nucleation in interphase plant cells, possibly through the recruitment of γ-TuRC. This is an important discovery that advances our understanding of the relationship between MT nucleation and cortical MT organization. The amiR-AUG6 lines appeared to have well aligned MT organization compared to wild type cells, consistent with reduced branching MT nucleation and more prevalent parallel MT nucleation. It would be interesting to test whether this phenomenon is attributed to failure to recruit γ-TuRC to the MT nucleation sites. This study opens up many other interesting questions. For example, six-fold reduction of nucleation frequency apparently did not affect the total microtubule mass in epidermal cells. How does the cell maintain its fine cortical MT network

email: sul38@psu.edu

under compromised MT nucleation conditions? How does the geometry of the pre-existing MT array organization affect MT nucleation events? Why does augmin knockdown result in a switch from predominant branching nucleation to parallel nucleation? Are there additional anchoring proteins required for the bridging between the augmin complex and γ-TuRC? Future work will be expected to address these eminent questions.

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